IJIR: Your Sexual Medicine Journal
https://doi.org/10.1038/s41443-021-00449-0

ARTICLE

Comparative assessment of outcomes and adverse effects using two different intramuscular testosterone therapy regimens: 100 mg IM weekly or 200 mg IM biweekly

Farouk M. El-Khatib[1] · Linda M. Huynh[1] · Alexei Kopelevich[1] · Mohamad M. Osman[1] · Edward Choi[1] · Jeanie T. Nguyen[1] · Sharmin Dianatnejad[1] · Qiaqia Wu[1] · Madeline G. Olivas[1] · Aaron Spitz[1] · Jacob Lowry[2] · Boriss Y. Losso[2] · Mohit Khera[2] · Laura Angulo-Llanos[3] · Premal Patel[3] · Ranjith Ramasamy[3] · Faysal A. Yafi[1]

Received: 14 September 2020 / Revised: 23 April 2021 / Accepted: 10 May 2021
© The Author(s), under exclusive licence to Springer Nature Limited 2021

Abstract

This study aimed to compare the change in levels of several laboratory values and the development of adverse events using two commonly used intramuscular testosterone therapy regimens. Men were included if they were 18 years or older and received one of the following testosterone therapy regimens: 100 mg intramuscular once weekly or 200 mg intramuscular once every other week. Primary outcomes were relative changes in total testosterone, free testosterone, estradiol, prostate-specific antigen, and hematocrit at 6 months after initiation of testosterone therapy. Secondary outcomes were any significant rises in estradiol, hematocrit, prostate-specific antigen, and any other treatment-related adverse events requiring cessation of testosterone therapy. A total of 263 men were enrolled. In a subanalysis of men who had a baseline hematocrit below 54% before intramuscular testosterone therapy initiation, we found the following: men who received 100 mg weekly injections were significantly less likely to have hematocrit levels rising above 54% (1/102 (1%) vs. 4/51 (8%); $p = 0.023$). No significant differences were recorded in the increase in total testosterone, free testosterone, prostate-specific antigen, and estradiol levels between both groups. A higher average serum testosterone over the dosing interval seen with the 200 mg regimen appears to be associated with a higher risk of erythrocytosis.

Introduction

Throughout the male lifespan, testosterone plays a critical role in sexual function. An increasing body of evidence suggests that testosterone also regulates multiple physiological processes, such as body development, bone metabolism, and hematopoiesis [1, 2]. The prevalence of hypogonadism varies amongst study populations and by geographical location, and has a reported range of about 12–38%, depending on the study groups chosen and on the methodology for measuring testosterone and cutoffs for testosterone level [3].

Testosterone therapy (TTh) aims to restore testosterone levels and reverse the signs and symptoms of hypogonadism. Testosterone can be delivered via injectables, topical, sub-dermal, intranasal, or oral routes [4]. Currently, intramuscular (IM) injectable testosterone esters (enanthate and cypionate) are amongst the most used formulations [5]. Testosterone cypionate are slow-acting esters with a release time between 8 and 10 days and are typically injected on a weekly or biweekly basis [6]. While the impact of testosterone administration on prostate-specific antigen (PSA) and prostate cancer (PCa) is debatable, multiple studies have shown that TTh does not increase PSA levels in men being treated for hypogonadism, except when it is given IM (very minimal increase), and based on evidence in the literature, TTh does not appear to increase the risk of PCa incidence among these men [7]. Another concern with TTh is secondary erythrocytosis and its potential impact on blood viscosity, which is most notable with injectables [8].

✉ Faysal A. Yafi
fyafi@hs.uci.edu

[1] Department of Urology, University of California, Irvine Health, Orange, CA, USA

[2] Department of Urology, Baylor College of Medicine Houston, Houston, TX, USA

[3] Department of Urology, University of Miami Health System in Florida, Miami, FL, USA

Published online: 13 July 2021

Although there is a growing body of research investigating the safety of TTh, including comparisons of different types of TTh administration, there are limited data comparing the efficacy and side effects of varying IM-delivered testosterone dosages. We seek to conduct a multi-institutional, retrospective chart review for men prescribed 100 mg IM weekly and men prescribed 200 mg IM every 2 weeks to assess (1) the relative changes in total testosterone (TT), free testosterone (FT), estradiol (E), PSA, and hematocrit (Hct) 6 months after initiation of TTh as our primary outcomes, and (2) significant rises in Hct, PSA, and E as our secondary outcomes. Testosterone cypionate (DEPO®-TESTOSTERONE|Pfizer | USA) was used for both TTh regimens.

Materials and methods

Study population

After obtaining institutional review board approval (IRB 2017-3746), we conducted a multi-institutional, retrospective chart review of men who presented to three different urology clinics between 2015 and 2019. Consented patients were all 18 years or older and received TTh either in 100 mg IM formulations once weekly or 200 mg IM formulations once every other week at any point during the 4 years. Patients were excluded from our data analysis if they received any additional non-TTh hormone replacement therapies (human chorionic gonadotropin, selective estrogen receptor modulators, and aromatase inhibitors) or if they had a current or previous PCa diagnosis.

Main outcome measures and data collection

Primary outcomes were absolute and relative changes in TT, FT, E, PSA, and Hct at 6 months after initiation of TTh. Secondary outcomes were any significant rises in E, Hct, and PSA defined as levels higher than 40 pg/mL, 54%, and 4 ng/mL, respectively. Any treatment-related adverse events requiring cessation of TTh were noted.

Levels for all parameters were obtained at baseline (before TTh initiation), and at 6 months after TTh initiation. Testosterone levels were most routinely checked mid-cycle at days 4–5 after the injection, or at days 7–8 after the injection for patients receiving 100 mg IM formulations once weekly or 200 mg IM formulations once every other week, respectively. The change in each parameter was compared to the baseline value, and the difference was calculated for each patient (labeled as delta). The average of the deltas was calculated and compared using the two-sample Student's *t*-test. Levene's test for the quality of variances was used to assess for parametric versus non-parametric distributions, and relevant *t*-tests for equality of means were utilized based on results for each variable. SPSS™ was used for data analysis, and all reported *p* values were two-sided, with $p < 0.05$ considered statistically significant.

Table 1 Baseline demographics.

	100 mg IM weekly ($N = 169$)		200 mg IM biweekly ($N = 94$)		*p* value
	N	%	*N*	%	
Diabetes	36/170	21.2%	10/83	12.0%	0.075
Hypertension	57/134	42.5%	38/84	45.2%	0.696
Dyslipidemia	25/110	22.7%	24/80	30.0%	0.257
Vascular disease	6/110	5.5%	9/80	11.2%	0.151
	Mean	SD	Mean	SD	
Age (years)	53.7	12.4	56.9	12.6	0.053
BMI (kg/m2)	30.1	5.0	30.4	5.4	0.657
HbA1c (mmol/mol)	5.9	0.7	6.6	1.6	0.068
Glucose (mEq/L)	106.0	30.8	107.0	23.8	0.889
Total Testosterone (ng/dL)	234.8	63.7	250.4	75.0	0.115
Free Testosterone (ng/dL)	37.6	24.1	44.6	18.8	0.114
Estradiol (pg/mL)	25.7	10.4	26.4	18.5	0.806
PSA (ng/mL)	1.4	1.7	1.1	1.3	0.271
Hematocrit (%)	45.1	4.8	46.3	4.1	0.110

Student's *t*-tests was used to compare averages.

IM intramuscular, *BMI* body mass index, *HbA1c* hemoglobin A1c, *PSA* prostate-specific antigen.

Results

Between 2015 and 2019, we identified a total of 263 men who had received IM TTh and complete 6 months follow-up data: 169 men received 100 mg IM once weekly, and 94 men received 200 mg IM once every other week. A summary of the baseline demographics can be found in Table 1 and the changes in different laboratory values are compared and reported in the graphs below (Fig. 1).

Testosterone

At 6 months, there was no significant difference in the change in TT levels (404.6 ng/dL for men on 100 mg IM once weekly vs. 460.4 ng/dL for 200 mg IM once every other week, $p = 0.555$) or FT levels (58.7 ng/dL for men on 100 mg IM once weekly vs. 92.1 ng/dL for 200 mg IM once every other week, $p = 0.362$). Overall, 68% of patients receiving 100 mg IM once weekly, and 67% of patients receiving 200 mg IM once every

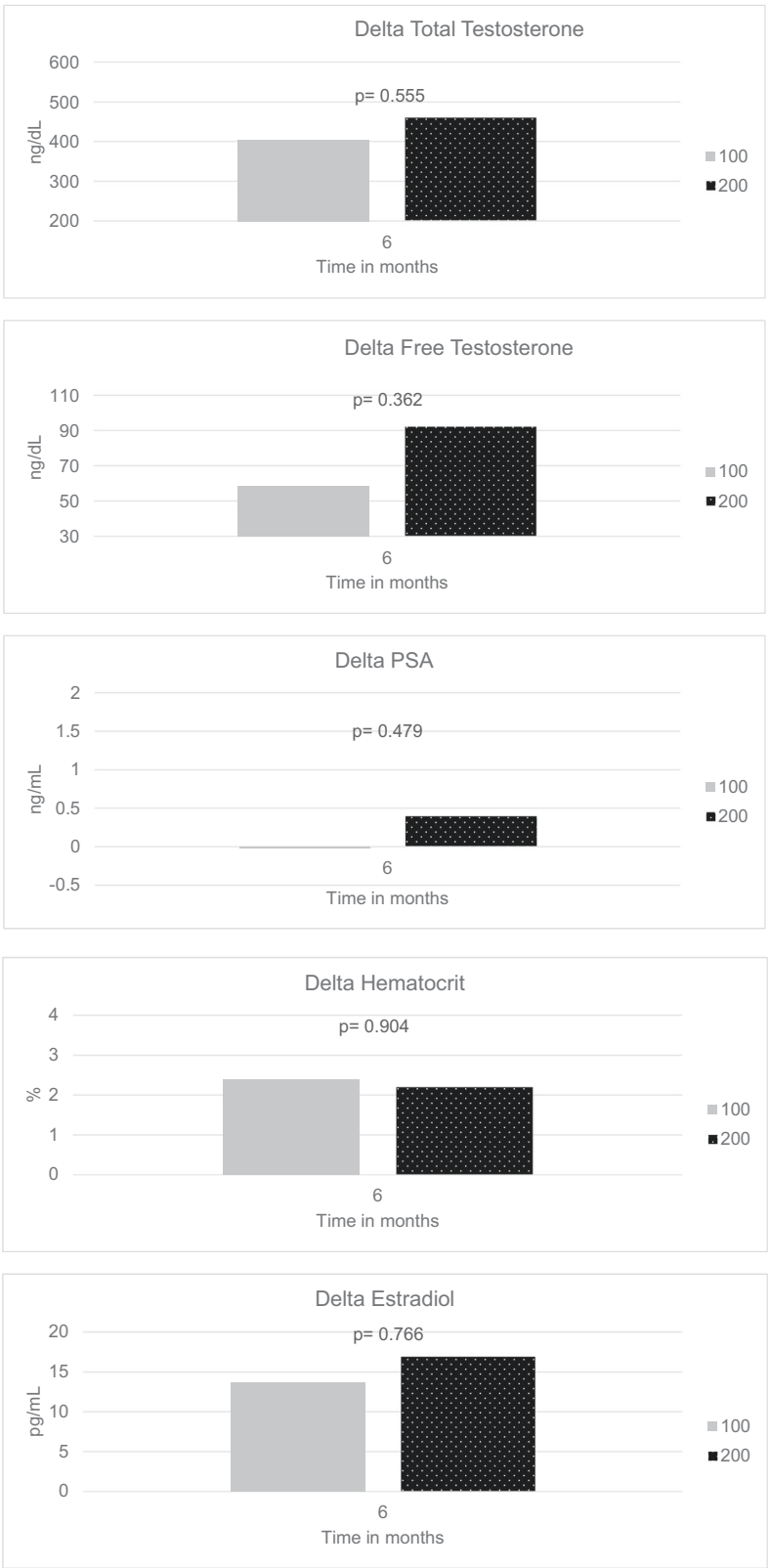

Fig. 1 The changes in laboratory values (delta) at six months after TTh initiation.

Table 2 Secondary outcomes.

$N = 263$	100 mg ($N = 169$)	200 mg ($n = 94$)	p value
Hematocrit > 54%	1/102 (1%)	4/51 (8%)	0.023
Estradiol > 40 pg/mL	8/48 (17%)	5/22 (23%)	0.552
Prostate Specific Antigen > 4 ng/mL	2/96 (2%)	1/42 (2%)	0.912
Aromatase inhibitor initiated	3/169 (2%)	4/94 (4%)	0.340
Phlebotomies performed	0	0	n/a

Explanation: the (one-sided) z test for equality of two percentages using independent samples was used to compare the different ratios.

other week were within the therapeutic range (TT 300–1000) ($p = 0.917$).

Hematocrit

At 6 months, there was no significant difference in the change in Hct levels (2.4% for men on 100 mg IM once weekly vs. 2.2% for 200 mg IM once every other week, $p = 0.904$). For the secondary outcomes, a subanalysis had to be performed. Out of the 169 patients who received 100 mg IM once weekly, 102 had a Hct value reported before TTh initiation and out of the 94 patients who received 200 mg IM once every other week, 51 had a Hct value reported before TTh initiation. All the 102 and 51 patients had a Hct baseline <54%. Of those receiving 100 mg IM once weekly, 1/102 (1%) had levels rising above 54%, and of those receiving 200 mg IM once every other week, 4/51 (8%) had levels rising above 54% ($p = 0.023$) (Table 2).

PSA

At 6 months, there was no significant difference in the change in PSA levels (−0.02 ng/mL for men on 100 mg IM once weekly vs. 0.42 ng/mL for 200 mg IM once every other week, $p = 0.479$). For the secondary outcomes, a subanalysis had to be performed. Out of the 169 patients who received 100 mg IM once weekly, 96 had a PSA value reported before TTh initiation and out of the 94 patients who received 200 mg IM once every other week, 42 had a PSA value reported before TTh initiation. All the 96 and 42 patients had a PSA baseline <4 ng/mL. Of those receiving 100 mg IM once weekly, 2/96 (2%) had levels rising above 4 ng/mL, and of those receiving 200 mg IM once every other week, 1/42 (2%) had levels rising above 4 ng/mL ($p = 0.912$) (Table 2). One patient among those receiving 200 mg IM every 2 weeks, underwent a prostate biopsy after his PSA rose from 2.2 ng/mL at baseline to 3.5 ng/mL at 6 months after TTh. Biopsy showed PCa with Gleason grade 1.

Estradiol

At 6 months, there was no significant difference in the change in E levels (13.6 pg/mL for men on 100 mg IM once weekly vs. 16.9 pg/mL for 200 mg IM once every other week, $p = 0.766$). For the secondary outcomes, a subanalysis had to be performed. Out of the 169 patients who received 100 mg IM once weekly, 48 had an E value reported before TTh initiation and out of the 94 patients who received 200 mg IM once every other week, 22 had an E value reported before TTh initiation. All the 48 and 22 patients had an E baseline <40 pg/mL. Of those receiving 100 mg IM once weekly, 8/48 (17%) had levels rising above 40 pg/mL, and of those receiving 200 mg IM once every other week, 5/22 (23%) had levels rising above 40 pg/mL ($p = 0.552$) (Table 2). Initiation of an aromatase inhibitor to suppress rising E levels was required in 2% of patients in those receiving 100 mg IM once weekly and 4% of patients in those receiving 200 mg IM every 2 weeks ($p = 0.340$).

Discussion

This multi-institutional retrospective study presents new data comparing the efficacy and safety profiles of two different IM TTh dosing regimens. The effects of both TTh, either in 100 mg IM formulations once weekly or 200 mg IM formulations once every other week, were mostly similar from a safety profile, but men receiving the 200 mg regimen were more likely to experience significant erythrocytosis.

The erythrogenic effect of testosterone has been well established. A recent systematic review and meta-analysis of randomized, placebo-controlled trials assessed the efficacy and adverse events of TTh for at least 12 weeks in adult men with hypogonadism [9]. In three studies ($n = 1579$), participants who received TTh experienced a significantly higher frequency of erythrocytosis (hematocrit >54% or hemoglobin >17.5 g/dL) compared with placebo (RR: 8.14; CI 95%, 1.87, 35.40) [9]. In theory, a significant increase in Hct may lead to thromboembolic events, likely due to higher blood viscosity and platelet adhesiveness and lower venous return [10]. The relationship between erythrocytosis and thromboembolic events is, however, mostly extrapolated from series examining patients with polycythemia rubra vera, a condition that is inherently prothrombotic due to a gene mutation [11, 12]. There is, however, still insufficient evidence supporting a direct relationship between TTh-induced erythrocytosis and cardiovascular or thromboembolic events. In a small sample ($n = 15$) retrospective study, Krauss et al. examined hypogonadal men receiving short-acting IM testosterone 300 mg

every 3 weeks [13]. Using a cutoff of 48%, they observed a correlation between strokes and transient ischemic attack incidence in men with Hct ≥48% (3/9 vs. 0/6; $p = 0.129$). Due to the small sample size and statistically nonsignificant results, the ability to draw conclusions from this study is small. The relationship between TTh and cardiovascular events as a whole remains an unanswered question, though most recent literature seems to demonstrate safety, and possibly protection. In a recent meta-analysis of randomized controlled trials related to TTh and cardiovascular risk, 73 trials reported information on major adverse cardiovascular events (MACE) [14]. The analysis showed that TTh was not associated with any significant difference in the incidence of MACE compared to placebo (OR: 1.01 [0.57;1.77]; $p = 0.980$). One major finding in our study was that men on the 200 mg biweekly regimen experienced a significantly higher rise in their Hct levels at 6 months after TTh initiation. This is likely because patients receiving a higher dose were more likely to spend a longer duration of time outside the normal reference range due to the higher average plasma concentration, which in turn may cause increased erythrocytosis.

In this study, only 2% of men in each group reached a PSA level higher than 4 ng/mL ($p = 0.912$). Concerns for testosterone and PCa arose in 1941 when Huggins and Hodges asserted that testosterone activated PCa and caused it to undergo an enhanced growth rate, which led to a general concern that higher serum concentrations pose a risk for PCa [15]. A few decades later, our understanding has evolved, and we now recognize that prostate tissue is sensitive to changes at low testosterone levels, yet indifferent at higher levels [16]. A recent meta-analysis looked at the association between testosterone, both endogenous and exogenous, PSA, and PCa [17]. Twenty-six placebo-controlled trials reported data on PSA levels, and 11 reported data on the incidence of PCa. In these 26 trials, the overall relative difference in PSA levels after the onset of use of TTh was only 0.10 ng/mL (−0.28, 0.48). Findings were similar in heterogeneity analyses by mode of administration, region, age at baseline, baseline testosterone, trial duration, type of patients, and type of TTh [17]. These findings show that although a rise in PSA can be observed in men on TTh, the risk of PCa remains the same with the appropriate monitoring.

Symptoms and signs of hyperestrogenism include gynecomastia, breast pain, and increase in visceral obesity, which can be detected in 10–25% of men on TTh [18]. There was no significant difference between the two regimens in our study regarding increases in E levels or a clinically significant rise in E (>40 pg/mL), nor were there any patients who complained of signs and symptoms of hyperestrogenism. In a retrospective review of charts of 34,016 men, Tan et al. wanted to examine the impact of aromatization on patients, and recorded a total of 7215 (20%) men with high E levels defined as ≥42.6 pg/mL [19]. These results are in line with the clinically meaningful increases in E noted in our study (17% and 23%, respectively). In their study, they also reported that high E levels were not associated with higher rates of low libido, but documented higher rates of low libido among patients with normal or lower E levels ($p < 0.05$) [19].

Our study is the first to show that there may be an advantage to administering smaller doses of IM TTh at a higher frequency, to minimize troughs and symptom return. In this regard, these results should be considered within the context of several limitations. This study was limited by its retrospective nature, lack of randomization and lack of power analysis for sample size assessment. Moreover, due to its multi-institutional nature, we could not control with high accuracy the timing of serum testing relative to testosterone injection, and the databases had only objective laboratory values and subjective qualitative entities related to side effects, which deprived us from comparing the clinical outcomes and hypogonadal symptoms between the two groups. TTh compliance was mainly done by asking patients, while a more objective approach would have been measuring luteinizing hormone levels since a value below 1 IU/L suggests compliancy. More prospective studies with longer follow-up are, however, needed to validate our findings further.

Conclusions

Adherence to a lower dosage, higher frequency IM TTh regimen seems to yield similar benefits in raising TT and FT levels and a significantly lower risk of having hematocrit levels rising above 54%. Future studies need to evaluate whether weekly TTh dosing would be associated with similar changes in symptomatology as compared to bi-monthly dosing.

Compliance with ethical standards

Conflict of interest The authors declare no competing interests.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

References

1. Gooren LJ, Bunck MC. Androgen replacement therapy: present and future. Drugs. 2004;64:1861–91. https://doi.org/10.2165/00003495-200464170-00002
2. Kaufman JM, Vermeulen A. The decline of androgen levels in elderly men and its clinical and therapeutic implications. Endocr Rev. 2005;26:833–76. https://doi.org/10.1210/er.2004-0013.
3. Zarotsky V, Huang M-Y, Carman W, Morgentaler A, Singhal PK, Coffin D, et al. Systematic literature review of the epidemiology

of nongenetic forms of hypogonadism in adult males. J Horm. 2014;2014:1–17.
4. Nieschlag E. Testosterone treatment comes of age: new options for hypogonadal men. Clin Endocrinol. 2006;65:275–81. https://doi.org/10.1111/j.1365-2265.2006.02618.x
5. Minnemann T, Schubert M, Freude S, Hübler D, Gouni-Berthold I, Schumann C, et al. Comparison of a new long-acting testosterone undecanoate formulation vs testosterone enanthate for intramuscular androgen therapy in male hypogonadism. J Endocrinol Investig. 2008;31:718–23. https://doi.org/10.1007/BF03346421
6. Hudson's guide: testosterone types and delivery. Hudson's FTM Resource Guide. http://www.ftmguide.org/ttypes.html#bib.
7. Kang D-Y, Li H-J. The effect of testosterone replacement therapy on prostate-specific antigen (PSA) levels in men being treated for hypogonadism: a systematic review and meta-analysis: a systematic review and meta-analysis. Medicine. 2015;94:e410.
8. Jones SD Jr, Dukovac T, Sangkum P, Yafi FA, Hellstrom WJG. Erythrocytosis and polycythemia secondary to testosterone replacement therapy in the aging male. Sex Med Rev. 2015;3:101–12.
9. Ponce OJ, Spencer-Bonilla G, Alvarez-Villalobos N, Serrano V, Singh-Ospina N, Rodriguez-Gutierrez R, et al. The efficacy and adverse events of testosterone replacement therapy in hypogonadal men: a systematic review and meta-analysis of randomized, placebo-controlled trials. J Clin Endocrinol Metab. 2018. https://doi.org/10.1210/jc.2018-00404.
10. Ohlander SJ, Varghese B, Pastuszak AW. Erythrocytosis following testosterone therapy. Sex Med Rev. 2018;6:77–85. https://doi.org/10.1016/j.sxmr.2017.04.001
11. Griesshammer M, Kiladjian J-J, Besses C. Thromboembolic events in polycythemia vera. Ann Hematol. 2019;98:1071–82.
12. Cuthbert D, Stein BL. Polycythemia vera-associated complications: pathogenesis, clinical manifestations, and effects on outcomes. J Blood Med. 2019;10:359–71.
13. Krauss DJ, Taub HA, Lantinga LJ, Dunsky MH, Kelly CM. Risks of blood volume changes in hypogonadal men treated with testosterone enanthate for erectile impotence. J Urol. 1991;146:1566–70. https://doi.org/10.1016/s0022-5347(17)38168-5
14. Corona G, Maseroli E, Rastrelli G, Isidori AM, Sforza A, Mannucci E, et al. Cardiovascular risk associated with testosterone-boosting medications: a systematic review and meta-analysis. Expert Opin Drug Saf. 2014;13:1327–51. https://doi.org/10.1517/14740338.2014.950653
15. Nelson WG. Commentary on Huggins and Hodges: "studies on prostatic cancer. Cancer Res. 2016;76:186–7.
16. Khera M, Crawford D, Morales A, Salonia A, Morgentaler A. A new era of testosterone and prostate cancer: from physiology to clinical implications. Eur Urol. 2014;65:115–23. https://doi.org/10.1016/j.eururo.2013.08.015
17. Boyle P, Koechlin A, Bota M, d'Onofrio A, Zaridze DG, Perrin P, et al. Endogenous and exogenous testosterone and the risk of prostate cancer and increased prostate-specific antigen (PSA) level: a meta-analysis. BJU Int. 2016;118:731–41. https://doi.org/10.1111/bju.13417
18. Eckman A, Dobs A. Drug-induced gynecomastia. Expert Opin Drug Saf. 2008;7:691–702. https://doi.org/10.1517/14740330802442382
19. Tan RS, Cook KR, Reilly WG. High estrogen in men after injectable testosterone therapy: the low T experience. Am J Mens Health. 2015;9:229–34. https://doi.org/10.1177/1557988314539000